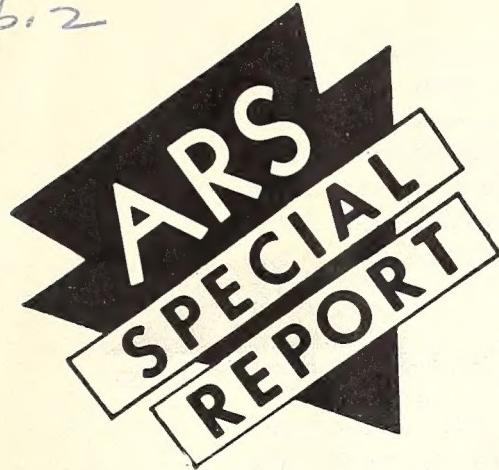


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✓ Use of Diseases To Kill Plant Insect Pests

A Research Progress Report

ARS 22-74



Growth Through Agricultural Progress

October 1961

Agricultural Research Service
UNITED STATES DEPARTMENT OF AGRICULTURE

FOREWORD

Killing destructive insects with their own diseases is a pest control method now used to some extent in the United States and other countries. The method has some distinctive advantages and is regarded as a worthwhile addition to other methods of pest control. Research is in progress to learn more about its merits for protecting forest trees and agricultural crops.

This report compiles information designed primarily to inform Extension and other agricultural leaders about research developments in microbial pest control--a method not very familiar to farmers or the general public.

Information in this report was provided by the Entomology Research Division of the Agricultural Research Service and the Division of Forest Insect Research of the Forest Service.

USE OF DISEASES TO KILL PLANT INSECT PESTS

A Research Progress Report

Man is learning, through scientific methods, to get more dependable benefit from a kind of pest control observed in nature: the killing of insects by their own diseases.

This control in nature is uncertain. Now and again, a pest-ridden forest or farm crop is rescued from destruction by a virus or other infectious disease organism that spreads and kills the ravaging insect. But many voracious insects wander or are carried about, and leave their fatal diseases behind. And when a pest does meet up with a disease agent, the kill of the pest is often too little or too late to be of much use to man. Over the years, nature's help with insect diseases falls short of what man may achieve by applying the right disease agents in a scientific way.

A scientific approach is to apply a disease organism in large quantities, in a spray or dust, at the proper time, with due regard for insect habits, weather, and other conditions on which a successful kill depends. In research with this method, scientists have employed varied pathogens (disease-causing organisms)--including bacteria, fungi, protozoa, viruses, and bacteria-carrying nematodes. As an informal word for any of the disease agents so used, many scientists accept the term "microbe." And the sprays and dusts containing microbial material are often called microbial insecticides.

A scientist's concept of the ideal microbial insecticide has been put in these terms: It should be highly infectious for at least one pest insect, preferably for a large number of kinds. It should be easily and inexpensively produced. It should be capable of being stored for a long period. It should pose no hazard to man, or to animals including beneficial insects.

Carefully tested microbes are now produced in research laboratories on a quantity scale and used under scientific guidance against some forest and farm pests in the United States and other countries. In addition, the public in this country can buy two bacterial insecticides commercially produced for uses designated on registered labels.

There is prospect of further uses for microbial materials through agricultural and forestry research under way. Interest in this use of insect diseases has been spurred by the resistance that many serious pests have been developing to chemical insecticides. Alternate chemicals have been introduced in some instances, and such adjustments will undoubtedly continue. At the same time, there is recognized need to gain command of varied pest-fighting tactics and weapons. Microbial materials also have possibilities for use in some situations where chemical residues would be objectionable.

Entomologists are hopeful that the microbial method can be used in protecting livestock and man against harmful insects, such as some flies and mosquitoes. Work toward this goal is under way by some research agencies, including those of the U. S. Department of Agriculture.

However, the most progress in making microbial insecticides practical has been achieved through research on protecting economic plants--the subject of this report.

WORLDWIDE RESEARCH, SHARED BY USDA

Worldwide interest in microbial crop protection is indicated by research reports that stem from all continents and some remote islands. More than 100 professional scientists are studying diseases of pest-type insects.

Especially important for progress are insect pathology laboratories, which are being established at an increasing rate. These laboratories commonly include pest control in their research programs.

The U. S. Department of Agriculture is among agencies that have pioneered in making the disease method of pest control practical. Present work is organized and shared along these lines:

The Entomology Research Division of the Agricultural Research Service maintains an insect pathology laboratory at the Agricultural Research Center in Beltsville, Md. In this laboratory, several scientists specialize in developmental work with insect disease agents as prospects for pest control. They investigate the most promising materials and methods of application in cooperation with the Entomology Research Division's specialists on pests of major farm crops. In addition, several ARS insect pathologists, stationed at other USDA laboratories, are working on field problems.

The USDA Forest Service conducts similar research on tree protection. Laboratory work centers primarily at Corvallis, Oreg.; New Haven, Conn.; and Beltsville, Md. Forest Service specialists team with ARS insect pathologists on field test projects.

And all of this USDA research is extended by cooperation with State agricultural experiment stations and other research agencies, including those in industry and in foreign countries.

THE MICROBIAL METHOD IN PERSPECTIVE

Pest Warfare Prospects

To understand how microbial insecticides fit into the overall situation of pest warfare, here are two premises on which scientific thinking and planning are based:

1. Man will need to improve and change his pest-fighting tactics progressively to have any hope of maintaining a good earth. The struggle will get harder, inevitably, as agriculture changes and as insects invade new areas through the expanding networks of transport and travel.

2. No single type of control is likely to give man an all-purpose pest destroyer. To combat different insects in different situations, man will need a variety of special-purpose weapons and methods. He will need to choose among these, and at times combine several, guided by the best recommendations that research can provide. Possibilities range from

chemical insecticides, cultural practices, and quarantines, to varied biological controls that include parasites, predators, and lethal diseases of pests.

Advantages of the Microbial Method

Merits of microbial control of insects are mainly these:

- A high kill of some serious pests can be achieved.
- Some microbial insecticides can be processed quickly for mass production, and some without elaborate equipment--a point of special interest in countries where chemical industry is not advanced.
- In some cases, a microbial insecticide applied once will persist and protect an agricultural locality for years. It may even spread its beneficial effect beyond the area treated.
- Usually a microbial insecticide and a chemical insecticide can be mixed and applied at the same time without interference, if combining them would give an advantage.
- Microbes that cause insect diseases tend to be so different from those attacking other forms of life that a selected kind need pose no hazard to man, livestock, wildlife, or plants. They offer a solution to some situations in which chemicals leave objectionable residues.

HOW THE METHOD EVOLVED

Early History

Though the fact that insects have diseases has been known for 2,000 years, killing pests with diseases could not be tried until the nineteenth century, when discoveries about disease organisms paved the way. As early as 1834, an Italian, Bassi, proved with silkworm experiments that a microbe could cause an infectious disease. By 1873, Le Conte in the United States first stated the proposition that insect diseases were worth careful study for pest control.

For a long time, efforts--in many countries--to introduce agents of insect diseases into pest-ridden localities met with frustration. Sometimes, a success was achieved but could not be repeated. The main reason for failures was a lack of exact knowledge about the relationships of a microbe to its insect host (an insect of a kind providing the microbe's requirements).

Clear-Cut Successes

From the 1930's on, dependable results began to come through research programs that provided every kind of working knowledge that researchers could think of. Three examples that are cited often are the bacterial control of the Japanese beetle; virus control of the European spruce sawfly; and the use of a virus and a bacterium, either one satisfactory, for killing the alfalfa caterpillar.

WORKING KNOWLEDGE FROM LABORATORY AND FIELD

Research on any microbial insecticide is detailed and complex. Here is a little information to indicate the kind of questions dealt with, and procedures and findings of the laboratory and field studies.

What Pathogens to Try Out

The field of search for microbial material is wide, since pathogens that infect insects number thousands. Many can be ruled out at the start because they cause only mild infections or are otherwise clearly unsuitable. Microbes worth testing have been found among bacteria, viruses, fungi, and nematodes. A bacteria-carrying nematode, known as DD-136, has shown that it can infect and kill many insect pests by introducing its bacterial load; and, in tests so far, this nematode has not been found harmful to plants, warm-blooded animals, or man. Recognized sources include also the protozoa and rickettsia, although on these little testing has been done.

How a Pathogen Works

Preliminary work includes taking note of symptoms that are often distinctive and useful in judging the killing rate of a disease. Signs of infected insects may be inability to feed, stunted growth, discoloration, paralysis. Disease-killed insects may hang in profusion on trees or other plants, or lie in masses on the ground, with characteristic appearance.

The laboratory specialist examines individual insects in stages of infection, to learn where, how, and at what speed an infection develops in an insect host. Organs, such as the digestive tract, are isolated for damage appraisal. Body tissues are microscopically examined. Usually, microbial infections kill by growing in the insect and breaking up tissue or by introducing substances toxic for the insect. Facts about the disease pattern are basic guides to dosages, timing, and other management of a prospective microbial insecticide.

What is the Infection Route

In insect life, two infection routes are most common. Many fungi produce spores that find their way onto a prospective host. These spores send out strong threadlike substances that can push through an egg shell, grub cuticle, or even the exo-skeleton that adult insects wear like armor. Nematodes also can pierce the body wall. Most of the bacteria, viruses, and protozoa cannot invade this way, and they are usually taken in with the insect's food. Establishing the main route is a guide to application. Some microbial materials can start a strong pest-killing attack only if applied to soil; others only if applied on plants.

How Timing, Temperature, and Moisture Influence Results

Temperatures at which insect pathogens can work differ greatly. Each pathogen and each insect has a range of temperatures that it can stand. Temperature testing determines the tolerances to heat and cold for a pathogen and insect host. Within these limits, the range at which infection

can occur is sometimes narrow. And generally, an optimum temperature is found, at which a pathogen launches its most effective attack.

Relative coolness at application time appears to favor the attack of a good many pathogens. In extensive studies with viruses at Beltsville, although a number of caterpillar pests could be infected at environments exceeding 100° F., both the symptoms and the death rates in these caterpillars increased rapidly as temperatures were dropped. Many bacteria and fungi multiply best at around 77° to 85° F. Some have failed to multiply at all in environments warmer than the low 90's or even 80's.

A low temperature requirement of a disease organism need not rule out its use in hot weather. Some larval hosts carry disease organisms down to cool levels in soil. Pathogens sprayed on plants such as trees often can be relied on to land in cool shade in quantities sufficient to do effective pest control work.

Moisture requirement tests have shown that some pathogens can stand as much dryness as their insect hosts, or more. Viruses have remained infective after severe tests in dry environments. However, humidity or rainfall generally aid the persistence and spread of insect pathogens, even those that can stand considerable dryness.

In order to grow in living tissue, any pathogen requires moisture, but once inside an insect host a pathogen has suitable moisture conditions automatically provided.

Time requirements are of prime importance for determining when to apply a microbial material, how soon to expect results, and how long the killing activity will continue.

An insect may prove susceptible to a pathogen only at one brief stage of its life cycle from egg to adult. In other cases, insects are found susceptible at all stages, although usually there is a time at which susceptibility is greatest.

The interval to expect between application of a microbe and an extensive pest kill can range from 24 hours to many days. Pathogens that develop slowly can be highly valuable when speed is not critical for saving a crop. Some viruses that work slowly, for example, have given outstanding protection against insect pests of tree crops.

Even chronic diseases that weaken but do not kill insects outright have possible usefulness in pest control, if the application is well-timed. Some protozoa interfere with normal egg production of host insects, thereby reducing prospective broods. And in a European field experiment, a protozoan applied to weevil-infested sugar beets weakened the weevil enough to permit a reduction in amounts of chemical insecticides ordinarily required for control.

What Equipment and Treatments Suit Field Uses

Since microbial sprays, dusts, and granules are similar in form to chemical insecticides, they can be applied with familiar hand or power ground equipment or aircraft used in agricultural and forest pest control. The USDA Forest Service has been giving special attention to improved air and ground equipment and techniques for applying microbial sprays.

For spraying, microbial materials are commonly diluted with water, and sometimes an ingredient such as corn sirup for a sticker is added. For solid or dry applications, microbial materials are mixed with a carrier, such as clay or talc, or with an insect bait, such as cornmeal.

Field testing of a microbial insecticide resembles field testing of a chemical insecticide. Specific examples dealing with microbial control of pests of forest trees and agricultural crops, are cited in later sections of this report.

Among general indications from field testing are these:

Some microbes that do not persist or spread have to be used like chemical insecticides. That is, a series of applications may be required to control a pest during crop production. Such use may give economic returns if the microbial material can be obtained at a reasonable price.

Many microbes that persist start pest-killing slowly but spread and build up in numbers. Some of these can persist in the soil; some in or on tissues of plants, such as trees; some only in host insects. Entomologists have reported protective action of certain of these pest-killers continuing for a number of years.

Safety in Use

Safety evidence comes from laboratory experiments with insects and vertebrates and cross-infection tests.

To start with, true insect diseases tend to be specific to insects. The viruses, bacteria, and other disease agents that kill insects generally are not capable of infecting other forms of life--from plants to man--because some requirement of the microbe is not met.

Body temperature of the host is one barrier. Some insect microbes can grow only at temperatures well below those of man and warm-blooded animals. Some of the sporeforming insect pathogens kill by developing toxic crystals that can grow only in insects.

Even when the laboratory specialist is satisfied that a microbe is no hazard to public safety, careful tests are made to try infecting other forms of life. Infection has never been reported from such tests either in plants or in animal life from man to earthworms. In reverse tests, insects generally have not proved susceptible to the classic diseases of man or animals--further evidence that the gap barring cross-infection is wide.

Furthermore, in natural outbreaks of insect diseases and in field experiments, the true insect diseases have a long record of limiting their activity to insects.

In any case, safety can be made doubly sure by concentrating on microbes that show a wide gap barring them from attacking valued life. Many insect diseases are so specific that it is possible to find kinds that kill a major pest yet are harmless to beneficial insects. Viruses are often limited to infecting a single insect species. Even pathogens that can kill a large number of species have no effect on other species.

The Federal Insecticide, Fungicide, and Rodenticide Act includes microbial materials among the products that must be registered by the U. S. Department of Agriculture before they can be sold interstate.

To obtain registration of a label, the manufacturer must prove that the material is effective and useful for the stated purpose and is safe when used as directed. Based on evidence submitted by the manufacturer, the U. S. Food and Drug Administration determines whether a residue tolerance should be established, or exemption from need for a tolerance should be granted. State laws that govern local sales usually are patterned after Federal law.

Man has made great progress in distinguishing the harmful from the harmless in microorganisms. Microbes of no importance to human health and safety are in fresh foods of all kinds and rightly taken for granted. Today's knowledge enables scientists to manage selected types of pest-killing microbes without adverse side effects.

MASS PRODUCTION

Microbial source materials are simple and go a long way. In University of California experiments, 5 virus-killed caterpillars were enough to spray an acre of caterpillar-infested alfalfa. Adding 5 dead caterpillars to 5 gallons of water gave each quarter-teaspoon of spray a virus strength of 5 million polyhedra (angular granules that surround virus particles and help protect them from becoming inactive). Each polyhedron contained several virus particles.

Two known ways of getting mass production are: (1) To collect or breed insects, infect them with a disease organism, and then put the disease-killed insects through a series of processes; or (2) to grow a disease organism outside its insect host in a culture medium, that is, using chemical nutrients.

The second way is preferable because it requires less work, and the processes and equipment are better adapted to economical commercial production.

Some fungi and bacteria can be grown in culture media, either in laboratory containers or in industry's vats of, say, 12,000-gallon capacity.

For the production of other insect disease organisms, infected insects are the only present sources. Efforts to culture these pathogens in chemical media are being made, but are still in exploratory stages. Typical difficulties encountered have been a scant production or failure of an organism to reproduce dependably after a generation or two.

Among the sporeforming bacteria that have failed so far to produce spores in practical numbers in artificial cultures are those causing milky diseases in Japanese beetle grubs. Experiments aimed at finding some satisfactory material on which these spores can be grown industrially have been started at the Fermentation Laboratory of the ARS Northern Utilization and Development Research Division, in Peoria, Ill.

Canadian Department of Agriculture scientists have indicated that they may attempt chemical rearing of nematodes, using techniques which they have developed for experiments in rearing insect parasites in various media.

Storage tests are included in production research, to learn the conditions in which a microbial material can be kept for future use and what storage life to expect. Some pathogens retain virulence well in storage; some do not. Many viruses that stand both dryness and cold can be air-dried and refrigerated, either in purified form or in dried larvae. Among viruses thus stored, some have remained virulent for 25 years. On the other hand, a virus used to kill the European spruce sawfly showed loss of virulence after 2 years in storage--indicating that keeping up fresh supply of this virus may be advisable, perhaps on a yearly basis.

COMMERCIAL PRODUCTS

At present, two kinds of microbial insecticides are produced commercially and registered with the USDA for specified uses. Here is general information on the commercial status and background of each.

A Japanese Beetle Killer

Preparations containing milky disease spores for killing Japanese beetle grubs are available to the public. When applied to soil or turf, the spores sicken and kill the grubs--the young of the beetle--where the grubs feed on plant roots underground. The preparations are sold under trade names through garden supply houses and similar outlets for soil or sod application by farmers, gardeners, and homeowners.

This spore material was developed and put to use years ago, and its history is one of the success stories of microbial pest control.

In Orient homelands, the Japanese beetle has never been a serious problem. But when it found its way to New Jersey, about 1916, the beetle seemed for a long time to have escaped natural enemies and it found some 275 kinds of trees, shrubs, field crops, and garden plants, and grass suited to its voracious appetite. Chemicals, quarantines, traps, and other controls curbed the beetle to some extent, but it crawled, flew, and ate its way into 14 Eastern States.

A disease was added to useful controls when, in the 1930's, some grubs of the beetle in New Jersey were found to be dying of a disease agent in the soil. A USDA research team identified two rather similar sporeforming bacteria. Both bacteria cause milky diseases, so named because the blood of the grubs, normally clear, turns milky white. A single disease-killed grub releases to the soil several billion milky disease spores. However, the disease in nature was found only in a small area. The fast-spreading beetle had mainly escaped this fatal disease.

The research team developed a method of getting quantity supplies of the spores by inoculating grubs and grinding up and otherwise processing the diseased insects. Government patents for producing this spore material were obtained in 1941 and 1942 by S. R. Dutky, a USDA leader in this research. Extensive tests showed that the spores do not harm man, domestic or wild animals or birds, or earthworms. The spores can kill only the Japanese beetle and a few related species.

In 1939, a systematic program of treating beetle-infested soil was started under guidance of USDA and State scientists, and by 1952 the spores were established in 14 Eastern States. Meanwhile, in the 1940's under

license from the Secretary of Agriculture, commercial companies were authorized to market the spore preparations to extend their usefulness.

Research showed that single treatments applied in spots a few feet apart in beetle-infested soil usually establish the spores. Infected grubs, moving about in the soil, spread the spores, and birds and animals that eat infested grubs or soil help to carry the spores to new areas. The process does not give dramatically quick relief from beetle damage, but in a season or two established spores show their underground effectiveness--Japanese beetle populations dwindle and foliage and turf damage is less.

Once well established, the spores tend to resist heat, cold, dryness, and moisture and are in the soil to stay for a long time, unless man strips away the soil layer containing them. Instances of the Japanese beetle becoming destructive in new suburban developments have been noted.

Where a fast kill of the grubs is important to save turf from serious damage, chemical insecticides continue to be useful. Also, chemicals may be needed for a fast kill of beetles when they are devouring foliage. Milky disease spores do not kill the adult insect.

A Killer of Many Pest Caterpillars

Preparations containing spores of a bacterium that scientists call Bacillus thuringiensis are being commercially produced for use on specified crops. Production has gone through the following stages:

Bacillus thuringiensis showed such promise for use against a number of caterpillar pests that several years ago agricultural specialists stressed a need for large supplies in ready-to-use form to permit field testing to expand.

In 1958, permits to several companies for shipment of preparations containing Bacillus thuringiensis were issued by the U. S. Department of Agriculture under the provision of the Federal Insecticide, Fungicide, and Rodenticide Act. A temporary exemption from the requirements of a tolerance was granted on about 50 food and feed crops, by the Food and Drug Administration. These actions made possible large scale testing by selected growers in continental United States, Hawaii, Puerto Rico, and Australia. The number of crops under temporary exemption has since been increased and this testing continues.

In the spring of 1960, the FDA established permanent exemptions from need for tolerances in using this material on 12 crops. The USDA has issued registrations for companies to market preparations containing the organism for control of certain caterpillars on some of these crops, namely, artichokes, broccoli, cabbage, cauliflower, celery, lettuce, potatoes; also on alfalfa for use limited to Arizona and the West Coast. In addition, a registration has been issued for use of the organism on tobacco, which requires no tolerance exemption.

No registration has been issued thus far for using the bacillus in control of tree pests. In both laboratory and field experiments, the bacillus has given encouraging results against a number of forest pests, but this research has not reached the stage of practical treatments being developed and recommended for use by timber growers.

Bacillus thuringiensis is one of the most versatile pathogens yet found in insect research. It kills more than 100 species of insects--notably caterpillars--that are economic problems, yet does not harm beneficial insects or other forms of life.

During spore production, the bacillus forms toxic crystals that act as a stomach poison in susceptible insects. When caterpillars feed where the spore material has been spread, some kinds die quickly, some more slowly, but all cease eating within minutes after feeding on treated plants. Some of these caterpillars are highly susceptible when young, and less so in their later larval life.

The spore material, including crystals, can be commercially produced in artificial media and does not lose virulence when the growth process is repeated. Moreover, the spores in dry storage retain their capacity for killing susceptible insects for 10 years, at least.

This bacillus has attracted researchers ever since its discovery in 1911 in Germany, where it was killing larvae of the Mediterranean flour moth. Up to World War II, European scientists had learned that the bacillus could kill larvae of the European corn borer, the gypsy moth, pink bollworm, and some other serious pests, but the war interrupted their experiments.

An American bacteriologist got a culture of the bacillus from Germany in 1936 and later sent some to the University of California where it was stored in a refrigerator as historic reference material for 7 years. In 1949, E. A. Steinhaus at the University tried the spores, found them still highly active, and experimented with them for killing the alfalfa caterpillar. To fight this pest, one of the most destructive in California, he had been working with a virus, first in the laboratory, later in field testing. Bacillus thuringiensis gave a faster kill than the virus. His practical and repeatable successes in spraying both of these microbial materials on caterpillar-ridden alfalfa in California attracted wide scientific attention.

Although a good deal is known about the bacillus, USDA scientists and others are still developing working knowledge for managing it in different field situations and weighing its value for specific uses.

FOREST PROTECTION

Insects on the rampage are the foremost cause of timber mortality, and forest entomologists are seeking economical ways to cut serious losses. Chemical sprays sometimes pay in emergencies, by giving a fast kill of certain pests. Microbial sprays on infested trees offer opportunity for lasting protection.

Since the United States and Canada have many tree problems in common, cooperation on microbial research to fight forest pests has been close, as some of the following examples indicate.

European Spruce Sawfly

The value of viruses in forest pest control was demonstrated first by Canadian Department of Agriculture scientists when the European spruce sawfly was wrecking thousands of acres of spruce in southeastern

Canada. The outbreak became evident in 1930 and tree destruction reached a peak by 1938. That year, spruce forests in Vermont and New Hampshire also were heavily infested. By 1940, the outbreak subsided, due to a disease that eventually brought about a tremendous kill. Canadian insect pathologists had identified the helpful agent as a virus. Laboratory tests showed that the virus was extremely virulent to the sawfly during its younger caterpillar stages. Infected larvae died in a week or two. In the spruce forests, the virus' victory came slowly, because for years the sawfly reproduced armies of hungry caterpillars faster than the virus could spread and kill.

Since 1940, the European spruce sawfly has been common in Canada and the Northeastern United States, but has not been a serious pest. If it should resume massive destruction, entomologists have a tested weapon--a virus spray developed in Canada that can be mass-produced and applied for a strong initial attack.

Two Pine Sawflies

Finding a virus deadly to a spruce sawfly encouraged entomologists to seek viruses for killing pine sawflies.

First success was attained with a virus that kills the European pine sawfly, a costly pest of pines in Canada and the United States. Canadian entomologists could find no virus foe of this pest in their part of the world, but they got one from Sweden in 1949. Using the gift--a few virus-killed larvae of the European pine sawfly--they developed a dilute spray and applied it successfully in 1950.

In 1951, they shared 100 gallons with the U. S. Department of Agriculture. Using this, Federal and New Jersey State scientists jointly tested treatments on Scotch and red pine infested with the European pine sawfly and got excellent results from either ground or air spraying. Ground spray tests, similarly successful, were made in Illinois by Federal forest entomologists and Illinois State forestry specialists.

This virus from Sweden has gained considerable practical use in Eastern and Central United States, in fighting the European pine sawfly under scientific guidance. The virus has not been commercially produced, but a bank of the virus extract from insect sources has been started recently at the Forest Insect Laboratory of the Forest Service, in New Haven, Conn. From experiments in Ohio, it is known that the virus in suspension can be stored at least 5 years without losing effectiveness. The bank enables the Forest Service to provide starter materials of the virus and a few other pathogens to research scientists, State pest control officials, and others who can mass-produce and process a supply. One virus spraying of trees infested with the European pine sawfly generally keeps this pest down for 6 or 7 years, provided the spray is applied at the right time in spring. A gallon of spray generally is enough for an acre and the strength allows for some pests to survive to enable the disease to persist.

A virus for killing the Virginia pine sawfly was discovered in Maryland in 1954, when USDA scientists were examining severely infested pine trees. They took dead larvae samples for disease tests, and found a virus in 30 larvae. Using this, they developed a spray and tried it at larval season in pinewoods infested with the Virginia pine sawfly. In a typical test, the spray killed about 77 percent of the larvae in 11 days.

Two Budworms

Efforts to control budworms with diseases have been underway for more than a decade, with partial success. The research has been focused mainly on the spruce budworm, which attacks our spruce and fir stands from coast to coast; and a close relative, the jack-pine budworm, which in the United States is mainly in the Great Lakes area.

Canadian scientists in 1949 found a protozoan disease at work in budworms in Ontario, and in 1955 started basic studies. Since effects of the chronic diseases caused by protozoa are exceptionally hard to trace, results of this work have been of particular interest. The Canadians got indications that the protozoan forms spores in the cells of spruce budworm larvae and these spores work two ways: (1) by multiplying and robbing the larvae of growth and energy; (2) by packing the mid-gut so that the larvae can digest less and less food. The spores worked fast enough to kill some larvae. These had no shrunken look of starvation. Laboratory analyses showed a breakdown of vital organs, and this seemed the likely cause of actual death. Infected female larvae that survived to adult stage were weakened and produced relatively few eggs.

A protozoan that infects the jack-pine budworm has been found also in Canada. It works very much the same way as the one infecting the spruce budworm, and its possibilities for use are also under study.

In the United States and in Canada, Bacillus thuringiensis has been found effective against the spruce budworm in some laboratory experiments.

Bark Beetles

When the worst forest pests of this country are mentioned, three types almost sure to be included are sawflies, budworms, and bark beetles. The bark beetles are hard to control and even hard to study. They are tiny and spend most of their lives hidden under bark. What diseases kill these pests in nature is only beginning to be learned. The Forest Service has begun systematic exploratory work to identify disease organisms that infect bark beetles in the West. Dead beetles collected from infested trees are analyzed. Pathogens found in specimens have included fungi, bacteria, and spirochetes. Bacillus thuringiensis is one bacterium that has been found infecting both the western pine beetle and the Engelmann spruce beetle.

Tent Caterpillars

Microbial insecticides show great promise for protecting trees and range browse from the tent caterpillars that spin tentlike webs. Experiments in this country have progressed farthest with the Great Basin tent caterpillar. This leaf-eater wrecks trees such as aspens and valuable browse plants such as bitterbrush in the West.

In one demonstration of what a virus can do, test areas of a Navajo Indian reservation in New Mexico were practically rid of the Great Basin tent caterpillar by spraying. To make this test, in 1957, the Forest Service and the California Agricultural Experiment Station sent an entomologist to the reservation. Working with the Indian tribe and the U. S. Bureau of

Indian Affairs, he sprayed a virus preparation that was processed from dead caterpillars over long test strips of aspens where tent caterpillar larvae were feeding. The virus gave a good initial kill, and showed that it could overwinter in the egg and give an even larger kill the following year. Laboratory studies indicated that the insect eggs play a significant role in this disease. The eggs can become infected from the outside, if virus spray lands on them. The eggs can also contain the virus initially, if laid by infected adults.

A different line of attack on the tent caterpillar has been opened up by encouraging tests with Bacillus thuringiensis. In one Forest Service experiment in Utah, serviceberry bushes infested with the Great Basin tent caterpillar were sprayed with this bacterial insecticide in late April. The four dosages tried gave kills of 90 percent or higher within a month. Dosages of the same strength in sun and in shade gave a higher kill where pests fed in the sun.

Gypsy Moth

Varied disease agents are being tested against larvae of the gypsy moth in a search for an efficient biological weapon suited to large-scale control operations. This pest, that is so destructive to forest and shade trees in the Eastern United States, had many natural parasite and disease enemies in Europe where it came from. In this country a virus that causes a wilt disease in the caterpillars has been the only disease agent known to launch effective attacks on gypsy moth infestations. ARS and Forest Service entomologists jointly have started laboratory and field tests on gypsy moth caterpillars with this virus and some other disease agents, including Bacillus thuringiensis and a bacteria-carrying nematode named DD-136.

FARM CROP PROTECTION

ARS entomologists who specialize in farm crop pests include varied microbes in field tests, but are focusing such work mainly on a few promising types. A major problem for growers of many of these crops is the control of destructive insects without an objectionable chemical residue. Some of the microbial field tests include comparison of killing rates with those of chemical insecticides in use.

The following examples indicate lines along which progress is encouraging. Some of this work is by ARS scientists alone, some in cooperation with State agencies and with industry.

European Corn Borer

Considerable disease work with the European corn borer is being done in Iowa, at the USDA Corn Borer Investigations Laboratory in Ankeny and cooperatively at the Iowa State University in Ames. A fungus (Beauveria bassiana), a bacterium (Bacillus thuringiensis), and a protozoan (Perezia pyraustae) are all considered good prospective help.

The fungus Beauveria bassiana is the one discovered in 1834 by Bassi and named for him. Spores of this fungus work particularly well against newly hatched larvae of first-brood corn borers, because these larvae

seek the whorl of the young corn plant as a favorite feeding place. Spores landing here at the right time find infant borers and favorable warmth and moisture. Timing for a high kill is critical because, as leaves open, the fungus spores are on the growing leaves and are carried away from the whorl. In field tests, the spores have usually been applied when 75 percent of the plants show leaf feeding. A second application 5 to 7 days after the first treatment has at times increased the effectiveness. The spores, for reasons that are obscure, are sometimes less effective against second-brood borers. Some of the factors that may be responsible are temperatures, the maturing corn, and other environmental conditions that are less favorable to the development of this disease in the insect. In some field tests, the spores have been disseminated with a cornmeal bait.

Evidence that the fungus spores can kill the European corn borer in its adult stage was obtained recently in experiments with male and female moths. In 5 days, all moths exposed to infection by the spores were dead; whereas untreated moths were all alive. This suggests that applications of the spores, rightly timed, might reduce egg laying.

The bacterium Bacillus thuringiensis has controlled the young borers as well as the fungus, or better. The bacillus spores have been tried in granules and in sprays. The granules worked better than a spray, apparently because the granules released spores in greater concentration in the whorl, where the borers accumulated.

The protozoan Perezia pyraustae, which is being studied for corn borer control, causes a chronic disease that kills some borers before they become adult and mate, and also lessens the capacity of female moths that survive to lay fertile eggs. This protozoan was first found attacking borers in American cornfields in 1949. It has since been reported in so many localities that it appears to be in nearly every corn borer horde. If its spores can be spread artificially, this protozoan may do more to reduce borer populations. Efforts to understand this protozoan are in early stages, and Iowa is one center for the research. Experiments have shown that the parasite protozoan can infect the borer at all stages, from egg to adult. A spore-counting technique, recently developed, has provided a helpful tool for managing dosages of the spores to get the maximum effect on borer larvae. In an individual borer, a heavy infection averages about 86 million spores; medium infection, 25 million; and light infection, 7 million.

Pink Bollworm

In research to protect cottonfields against the pink bollworm, microbes of several types have shown promise. Investigation of microbial materials is included in a USDA-State-industry program organized to search intensively for more effective and economical means of controlling this formidable pest. In preliminary trials at the USDA Pink Bollworm Laboratory in Brownsville, Tex., six fungi, nematode DD-136, and Bacillus thuringiensis all were found highly destructive to larvae of the pink bollworm. Progress toward practical use has advanced furthest with the bacillus, which is being tested on the pink bollworm to learn the temperatures and other environmental conditions that promote the most effective kill, and to compare the merits of soil and plant applications.

Tobacco Hornworms and Budworms

Applying dried spores of Bacillus thuringiensis in a cornmeal bait was tried in 1960 and found effective for killing budworms as well as hornworms that damage tobacco. In earlier experiments, sprays and dusts containing the spores killed hornworms, but gave only partial control of budworms. Since budworms and hornworms attack tobacco at the same time in the South, there is hope of adjusting dosage rates to get a combined kill and avoid foliage damage.

Adding cornmeal to spore dust preparations apparently induces budworm larvae to eat avidly enough to take in spores for a high infection rate. The spores do not kill at once, but worms that eat the spore-cornmeal bait soon stop feeding and die. In limited experiments, the effectiveness of the bait has equaled or exceeded the excellent control that growers obtain with chemical insecticide sprays containing endrin.

These experiments have been conducted on flue-cured tobacco in North Carolina, South Carolina, and Florida. In Florida, work has been done on both shade-grown and sun-grown tobacco. The spore bait is effective for several days, and in a typical experiment four treatments have been spaced over 2-week intervals. Hand dusters have been used, but the best results have been achieved when bait was applied to buds and upper leaves of tobacco plants by hand.

Caterpillar Pests of Leafy Vegetables

Bacillus thuringiensis is known to infect and kill many kinds of caterpillars that damage leafy vegetable crops. Pests susceptible to the bacillus spores include the cabbage looper, salt-marsh caterpillar, imported cabbage worm, beet army worm, and diamond-back moth. In some field tests, the bacillus spores have given an unexpectedly light kill, indicating that conditions favorable to the spores are still not thoroughly understood.

Among recent findings is evidence that the spores kill these leaf-eating caterpillars by two-way action of infection and starvation. In field cages in Arizona, for example, the bacillus spores in a dust application halted leaf damage of large and active salt-marsh caterpillars, so that scarcely a trace of feeding damage occurred for 8 days.

A virus that controls the cabbage looper at times by natural spread in crop areas has been studied for a number of years and is considered helpful. Quantities of this microbial material have been applied for crop protection by scientists. Like other viruses, this one can be mass-produced at present only by using living insects. Finding a way to culture it on a medium would bring this virus nearer to commercial production.

Since some caterpillars that damage leafy vegetable crops are susceptible to this virus and also to Bacillus thuringiensis, ARS scientists have made preliminary tests of mixing the two pathogen materials. If dosages can be adjusted, the possibility is seen that a mixed treatment might give a higher killing rate of some pests and at the same time kill more kinds of pests than either pathogen applied singly. Mixing pathogens with chemical insecticides is also being tried out for the same advantages in fighting leaf-eating caterpillars.

Citrus Red Mite

Finding a virus disease that kills the citrus red mite has been called a break for pest control research, because it is the first evidence that viruses can attack a member of the spidermite group.

ARS scientists stationed at the USDA laboratory in Whittier, Calif., discovered this disease in 1958, when they were studying the biology of the citrus red mite and found that a strange infection was killing off laboratory specimens. They collected and observed citrus red mites from southern California areas of the coast and interior, and some of these lots were virtually wiped out by the same disease in the first brood and several succeeding broods.

Suspecting a virus, the pathologists ground up diseased mites, passed the material through filter paper, and diluted the filtered substance to make a spray. This spray on healthy mite colonies caused infection and death in 7 to 18 days, and appeared lethal to all stages of the mites' life cycle except the egg. A scientist at Cambridge, England, confirmed the virus nature of the disease by studying diseased mites with an electron microscope. Since discovery, the virus has been found in nature in scattered citrus groves of southern California.

In recent field tests in California, the method of applying the virus in a spray has been compared with introducing infected live mites into tree plots already mite-infested. This work is in preliminary stages.

Much remains to be learned about the mite-killing virus and its agricultural value. ARS scientists have developed improved methods of producing the virus in materials suited to field experiments and have supplied some to other researchers.

Scientists are interested in testing this virus on additional species of mites, because many of these pests have been developing resistance to chemical miticides. If the virus does not prove versatile for mite control uses, it has, in any case, alerted pest control workers to look out for other viruses that may be doing protective work in mite-infested crops.

LOOKING AHEAD

Insect pathologists regard their scientific specialty as a young one, but rapidly coming of age. In pest control work, they look forward to advances along lines indicated in this report. Finding ways to culture pathogens such as viruses, protozoa, and bacteria-carrying nematodes would be particularly useful.

There is prospect that the research will extend to new fields. For example:

Chemists and toxicologists can contribute notable assistance by determining the chemical properties of a toxic crystal, such as that produced by Bacillus thuringiensis. More exact knowledge of such crystals would go to the root of understanding the diseases, and could lead to short-cut ways of applying crystals directly, and possibly to synthetic production.

And while pathogens have been used in pest control almost entirely as insecticide materials, the possibility of using them another way is foreseen. Spreading live infected insects may prove a superior technique with diseases that can be transmitted through mating. Quantities of infected male adult insects released to seek their kind could reduce pest populations, even among insect species that mate only once. It is known that many pathogens lessen or destroy the egg-laying capacity of female insects. In some cases, the infection can be transferred via eggs to larvae. Furthermore, if a pathogen is a type that persists on plants or in soil, the infected insects would establish reservoirs of the disease agent by this means. USDA scientists have started exploratory work in this direction. Success with the infected insect method would give even better control than releasing sterile male insects, especially among species in which male insects mate several times.

